## OCT 0 2 2001 TECH CENTER 1600/2900

## What is claimed is:

1. (Amended) An immunoassay for detecting exposure to *Leishmania* parasites in a subject comprising the steps of:

contacting a sample from the subject suspected of having leishmaniasis with a soluble antigen prepared by [utilizing] <u>culturing the Leishmania parasites in</u> a protein-free medium <u>comprising an oncotic agent;</u> and

detecting the presence or measuring the amount of an antibody or fragment thereof in the sample bound to the soluble antigen.

- 3. (Amended) The immunoassay of claim 1, wherein the protein-free medium further comprises at least one of the following ingredients: Hepes buffer, L-glutamine and sodium bicarbonate without phenol red.
- 4. (Amended) The immunoassay of claim 1, wherein the antibody is IgG or IgM and is specific for a Leishmania antigen.
  - 5. (Amended) The immunoassay of claim 1, wherein the sample is a serum sample.
- 6. (Amended) The immunoassay of claim 5, wherein the serum sample is modified by diluting it 1:1000 in blocking buffer having 1.0% boiled casein.
- 7. (Amended) The immunoassay of claim 1, wherein said immunoassay is capable of diagnosing visceral, cutaneous or canine leishmaniasis in a subject.
- 8. (Amended) The immunoassay of claim 1, wherein the *Leishmania* [soluble antigen preparation is prepared by using] <u>parasites are</u> clones of *Leishmania donovani*, [or] *Leishmania mexicana*, or a combination thereof.

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11. (Amended) [A] The kit [for the diagnosis of leishmaniasis in a subject comprising a substrate and a soluble antigen] of claim 45, wherein the soluble antigen is of either L. donovani or L. mexicana [prepared by utilizing a protein-free medium comprising an oncotic agent packaged together for multiple or single use assays].

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- 12. (Amended) The kit of claim [11] <u>45</u>, wherein the substrate is coated with the soluble antigen.
  - 13. (Amended) The kit of claim [11] 45, further comprising a positive control.
  - 14. (Amended) The kit of claim [11] 45, further comprising a negative control.
  - 15. (Amended) The kit of claim [11] 45, further comprising a diluent.
- 16. (Amended) The kit of claim [11] <u>45</u>, further comprising an anti-human IgG conjugated to a label.
  - 17. (Amended) The kit of claim [11] 45, further comprising a substrate chromogen.
  - 18. (Amended) The kit of claim [11] 45, further comprising a substrate buffer.
  - 19. (Amended) The kit of claim [11] 45, further comprising a blocking buffer.
  - 20. (Amended) The kit of claim [11] 45, further comprising a stopping solution.
  - 27. (Amended) The kit of claim [11] 45, further comprising instructions.

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29. (Amended) A diagnostic device comprising a *Leishmania* soluble antigen prepared by [utilizing] <u>culturing a *Leishmania* parasite in a protein-free medium comprising an oncotic agent</u> and a means for detecting an antibody bound to the *Leishmania* soluble antigen.